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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.
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ART UNIT	**PAPER NUMBER**
1642	9

DATE MAILED: 04/06/01

Please find below and/or attached an Office communication concerning this application or proceeding.

Commissioner of Patents and Trademarks

Office Action Summary	Application No.	Applicant(s)	
	09/653,961	WU, GUANG-JER	
	Examiner	Art Unit	
	Stephen L. Rawlings, Ph.D.	1642	

-- Th MAILING DATE of this communication appears on the cover sh et with the correspond nce addr ss --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136 (a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *16 March 2001* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-19* is/are pending in the application.

4a) Of the above claim(s) *14-19* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-13* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claims _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. § 119

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☒ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

Attachment(s)

15) ☒ Notice of References Cited (PTO-892)
16) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
17) ☒ Information Disclosure Statement(s) (PTO-1449) Paper No(s) *3* .
18) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
19) ☐ Notice of Informal Patent Application (PTO-152)
20) ☐ Other: .

DETAILED ACTION

1. The Election filed on March 16, 2001 in Paper No. 5 of Group I (claims 1-6 and 12) with traverse is acknowledged and has been entered.

2. Claims 1-19 are pending in the application. Claims 14-19 have been withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in Paper No. 5.

3. Claims 1-13 are currently under prosecution.

Election/Restrictions

4. Applicant's election with traverse of Group I in Paper No. 5 is acknowledged. The grounds for traversal are that Groups I and IV are similarly classified, that Groups II and III are directed to the detection of the expression of the same protein as detected in Group I, and that searching all of the inventions would not be a substantial burden to the examiner. This argument is not persuasive. MPEP 802.01 provides that restriction is proper between inventions that are independent or distinct. With regard to Groups I and IV, the inventions are distinct for the reasons set forth in the previous Office Action mailed on January 18, 2001 (Paper No. 4). Additionally, classification of subject matter is merely one indication of the burdensome nature of the search required to examine the claims. While drawn to related subject matter, the inventions in Groups I and IV are divergent. The literature search, which is particularly relevant to this art, is not coextensive. Because the inventions are divergent, different searches are required to examine each group, which places a substantial burden upon the examiner. For these reasons, with regard to Groups I and IV, the restriction requirement is still deemed to be proper and is therefore made final.

After reconsideration of the restriction requirement, however, Groups II and III are rejoined with Group I, which now consists of claims 1-12.

With regard to Group V, because applicant did not distinctly and specifically point out the supposed errors in the restriction requirement, the requirement is deemed proper and is therefore made final.

Claim Rejections - 35 USC § 112

5. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

6. Claims 1-12 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method of identifying the metastatic potential of a prostate cancer cell line that expresses the gene encoding MUC18 (SEQ ID NO: 2), does not reasonably provide enablement for a method of identifying the metastatic potential of *any* prostate cancer cell that does not express the gene encoding MUC18 (SEQ ID NO: 2). The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims.

The claims are drawn to a method for determining whether a prostate cancer cell has the potential to metastasize, wherein said method comprises determining the level of expression of the gene encoding MUC18 in the prostate cancer cell and a normal prostate cell, comparing the levels of expression, and when the level of expression in the prostate cancer cell is higher than the level of expression in the normal prostate cell, identifying the prostate cancer cell as having metastatic potential.

The specification teaches that MUC18 is a glycoprotein of about 113 kilodaltons, which is variously recognized by other names and the presence of which has been demonstrated to be an important determinant of the ability of cancer cells, namely melanoma cells to metastasize (page 2, lines 6-16). The specification teaches that the gene encoding MUC18 is expressed in the metastatic PC-3, DU-145, and TSU-PR-1 prostate cancer cell lines, as evidenced by a Northern blot analysis. On the other hand, the specification teaches that the gene encoding MUC18 is not expressed in non-

metastatic LNCaP prostate cancer cell line (see page 14, Table 2 and Figure 2). The specification teaches that a Western blot analysis of protein lysates isolated from these cell lines also suggests that the gene encoding MUC18 is not expressed in LNCaP, but is expressed in PC-3, DU-145, and TSU-PR-1 (page 12, lines 17-25 and Figure 6). The specification also teaches that while a normal prostate epithelial cell line does not express the gene encoding MUC18, prostate cancer cells that were isolated from a patient biopsy have been found by Western blot analysis to express the gene (page 5, lines 7-14 and Figure 7). The specification discloses that the level of expression of the gene encoding MUC18 can be determined using conventional methodology, namely Northern blot analysis, quantitative RT-PCR analysis, and immunoassays (page 3, lines 3-14). In Example 1, the specification teaches a method of making chicken polyclonal antibodies that are capable of specific binding to the middle portion of the MUC18 polypeptide, which consists of the fragment spanning positions 211-376 of the amino acid sequence set forth in SEQ ID NO: 2 (pages 16-18). In Example 2, the specification identifies conventional techniques that can be used to determine the degree of motility and the invasiveness of prostate cancer cells (pages 18-19). In Example 3, the specification teaches that the expression of the MUC18 gene can be measured by Northern blot and quantitative RT-PCR analyses (pages 19-21). Finally, in Example 4, the specification teaches that immunofluorescence assays can be used to identify prostate cells that display MUC18 polypeptide at the cell surface (pages 21-22).

The teachings of the specification cannot be extrapolated to the enablement of the invention commensurate in scope with the claims, because the method cannot be used to identify a prostate cancer cell with metastatic potential that does not express the gene encoding MUC18. There are clear indications in the art that not all metastatic cancer cells express MUC18, and if this is so, the claimed method cannot always be used to identify the metastatic potential of all types of cancer. However, the claims are drawn specifically to the identification of the metastatic potential of the prostate cancer cells. Nevertheless, there is a problem: the art suggests that, regardless of the type of cancer, there is an uncertainty that every tumor biopsy from each different patient, though diagnosed with the same type of metastatic cancer, will test positive for MUC18

expression. As a consequence, the results of an analysis according to the claimed method may be highly unreliable.

It is reasonably well established that measuring the level of expression of MUC18 in malignant melanoma has prognostic value (see, for example, Luca, et al Form-PTO1449, citation G). However, Filshie, et al (*Leukemia* **12**: 414-421, 1998) analyzed the expression of the gene encoding MUC18 in several human leukemic cell lines and bone marrow sample from patients diagnosed with metastatic cancer. Filshie, et al teach that only one of five B lineage metastatic cell lines and one of four myeloid metastatic cell lines express MUC18 (abstract). Further, Filshie, et al teach that MUC18 positivity was observed only in 20% of B lineage acute lymphoblastic leukemia (ALL) patients, but all of the patients, of course, had malignant cancer (abstract). Similarly, only a fraction of patients diagnosed with malignant T lymphocyte (T)-ALL and a fraction of patients diagnosed with malignant acute myelogenous leukemia (AML) had cancer cells that tested positive for the expression of the gene encoding MUC18 (abstract). As another example, Shih, et al (*Clinical Cancer Research* **2**: 569-575, 1996) teaches that many formalin-fixed tissue samples biopsied from patients diagnosed with metastatic cancer do not express MUC18. Shih, et al disclose that neither fibrosarcomas, synovial sarcomas, or liposarcomas expressed MUC18 and only 15% of the malignant fibrous histiocytomas expressed the marker (abstract). Thus, there is obviously not a fore going assumption that metastatic cancer will express MUC18. More importantly, with regard to the instant application, it can not be presumed that, if a cancer cell does not express MUC18 it does not have the potential to metastasize, or even that it is not already metastatic. The teachings of Filshie, et al and Shih, et al indicate that MUC18-positivity cannot be used as the sole criterion for establishing whether or not a cancer cell has the potential to metastasize. While Filshie, et al and Shih, et al do not specifically teach that there is heterogeneous expression of MUC18 in a population of patients diagnosed with metastatic prostate cancer, *per se*, the implications of the teachings are still considered to be fully relevant to issues of enablement in the instant application. There is, at least, an inference that one skilled in the art cannot predict that just because one sample isolated from a patient is positive for MUC18 that all samples

from all patients diagnosed with metastatic prostate cancer will test positive. Moreover, there is potentially conflictive data in the art that may suggest that the teachings of the specification are inaccurate. US Patent 6,184,043 B1 teaches that the metastatic prostate cancer cell line DU-145 does not express the MUC18 polypeptide (Table 2a). This discrepancy suggests a need for further experimentation. It is noted that apart from the analysis of four prostate cancer cell lines, only one of which is not metastatic, the specification teaches the result of an analysis of a single patient biopsy. The number of samples isolated from patients that have been analyzed is certainly not sufficient to establish clinical or experimental significance, and it is not immediately apparent that that the claimed method can be used effectively in a clinical setting to determine whether a prostate cancer cell isolated from a patient has metastatic potential, in order to design a course of therapy appropriate for the patient. Consequently, one skilled in the art cannot practice the invention in a clinical setting without further undue experimentation to determine if, indeed, there is experimental and clinical significance, which would provide the artisan with a reasonable expectation of success.

It is well known in the art that tumors are heterogeneous conglomerates of cancerous cells, differentially expressing various tumor-associated antigens or markers, such as MUC18 and ErbB2. The lack of homogenous expression of tumor-associated antigens by tumor cells is frequently indicated to be the cause of failures to accurately diagnose and treat cancer in the art. In the case of MUC18, even normal endothelial tissues stain diffusely with anti-MUC18 antibodies, which is indicative of heterogeneous MUC18 expression by endothelial cells. Cell lines, such as PC-3, DU-145, and LNCaP, are used experimentally because they are generally homogeneous, but it is also appreciated in the art that the results of studies using cell lines are often not correlative with the results observed when primary cells are used in the same experiments. Again, it is noted that the specification demonstrates the expression of the gene encoding MUC18 in only one sample of primary origin, a tumor biopsy from a patient; otherwise, analyses of cell lines were used to support the premise that the expression of MUC18 is a determinant of metastatic potential in prostate cancer. Furthermore, it is noted that

the one non-malignant prostate cancer cell line (i.e., LNCaP) that was used in the study, which is presented here in this application, expresses a substantially higher level of E-cadherin than the other cell lines tested (see page 14, Table 2). Similarly to the expression of MUC18, E-cadherin expression also has been suggested to determine the degree of cancer cell motility and invasiveness, properties which define metastatic potential. Hsu, et al teach that "in a skin reconstruction model, ectopic E-cadherin expression inhibits invasion of melanoma cells into dermis by down regulating invasion-related adhesion receptors, MelCAM/MUC18 and beta3 integrin subunit, and by induction of apoptosis" (abstract). Therefore, as the specification teaches, because LNCaP expresses E-cadherin, it is not immediately apparent that the lack of MUC18 is the sole cause of the LNCaP cell line's inability to metastasize in nude mice; obviously there are other factors that contribute to the development of metastatic phenotype. Certainly, in view of the biologic complexity of gene expression that leads to the development of the metastatic phenotype in cancer cells, the demonstration in the specification that a single cell line that lacks MUC18 is not metastatic is insufficient to prove that the invention is enabled. The specification, however, is silent with regard to this issue.

Additionally, because a variety of normal tissues, besides the prostate express detectable levels of MUC18, it is clear that there will be complications resulting from the presence of these cells in biologic samples when practicing the invention. For example, Weninger, et al (*Journal of Investigative Dermatology* **115**: 219-224, 2000) teach that the gene encoding MUC18 is expressed in breast epithelia, hair follicles, and keratinocytes (i.e., the skin) (abstract). Shih, et al (Form-PTO 1449, citation L) teach that the gene is also expressed in vascular endothelium, smooth muscle, activated T cells, and intermediate trophoblast (page 745, column 1). Filshie, et al (cited supra) teaches that bone marrow fibroblasts express MUC18, which may be of particular significance in light of the fact that many prostatic cancer metastases may form in the bone. Moreover, Johnson, et al (Form PTO-1449, citation E) disclose that "in contrast to the restricted expression observed in cell lines, northern analysis of various normal human and murine tissues indicate that MUC18 expression in vivo is ubiquitous" (page

101, paragraph 2). It is well appreciated in the art that the presence of contaminating cells that express tumor-associated markers in samples acquired from tumor biopsies frequently skew the data to such a significant degree that the results of the studies are useless, or at best inconclusive. This problem is especially prominent in the context of a method that relies on data generated using the extremely sensitive method of reverse transcriptase PCR (RT-PCR). Obviously, meticulous micro-dissection is required to remove any and all histologically normal tissue when procuring and isolating cells from biopsied specimens, if the invention is to be practiced with a reasonable expectation of success. Yet, the specification provides no guidance with regard to this issue.

Finally, in view of the fact that Johnson, et al (cited supra) teach that the gene encoding MUC18 is ubiquitously expressed, one skilled in the art cannot practice the invention without an expression level index, which serves to instruct the skilled artisan what cut-off value for the level of expression delineates metastatic potential from non-metastatic potential. Clearly, the claims are drawn to a quantitative analysis of the level of MUC18 coding sequence expression. However, the specification provides insufficient guidance with regard to issue and most particularly, does not provide the expression level index that is essential to one's ability to successfully practice the invention.

In view of the above and particularly in the absence of sufficient guidance with regard to these issues and because of a lack of exemplification that is commensurate in scope to the claims, one skilled in the art cannot practice the invention, when drawn to the full breadth of the claims, with a reasonable expectation of success without undue experimentation.

7. Claims 1-5 are further rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method of identifying the metastatic potential of a prostate cancer cell line that expresses the gene encoding the MUC18 polypeptide (SEQ ID NO: 2) using an antibody capable of specific binding to the middle portion of the MUC18 polypeptide (SEQ ID NO: 2), which spans amino acid residues 631 to 1128 of SEQ ID NO: 2, does not reasonably provide enablement for a method of identifying

the metastatic potential of a prostate cancer cell that does not express the gene encoding MUC18 using *any* antibody made in an experimental laboratory animal in response to a MUC18 antigen. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention commensurate in scope with these claims.

The claims are drawn to a method for determining whether a prostate cancer cell has the potential to metastasize, wherein said method comprises determining the level of expression of the gene encoding MUC18 in prostate cancer cells and normal prostate cells by an immunoassay using *any* antibody made in an experimental laboratory animal in response to a MUC18 antigen.

The specification teaches what is set forth in the 35 USC § 112, first paragraph rejection above. The specification also discloses that a chicken polyclonal antiserum, which is immunospecific for the middle fragment of the MUC18 polypeptide that spans positions 211-376 of the amino acid sequence set forth in SEQ ID NO: 2, can be used to measure the level of expression of the gene encoding MUC18 in prostate cells in Western blot analysis (page 4, line 27 to page 5, line 14).

The teachings of the specification cannot be extrapolated to the enablement of the invention commensurate in scope with the claims, because the method cannot be practiced with any antibody made in an experimental laboratory animal in response to a MUC18 antigen. For example, Kraus, et al (*Melanoma Research* **7**: s75-81, 1997) teach that four anti-MUC18 antibodies, which were tested on frozen sections of melanoma metastases, had variable binding specificity. One of the anti-MUC18 antibodies (MUC18BA.3) reacted strongly with all lesions, perhaps indicating cross-reactivity with another melanocyte marker. In view of this result, it is apparent that not *all* antibodies made in an experimental laboratory animal in response to a MUC18 antigen can be used to distinguish metastatic cancer cells from non-metastatic cancer cells. Moreover, it is also apparent that a misdiagnosis could have very serious repercussion, so much so that any non-working embodiments of the invention may be considered to be unacceptably hazardous. US Patent No. 6,184,043 B1 teaches a method for detecting cells that express specific tumor-associated antigens using antibodies that are made in

laboratory animals in response to the antigens (abstract). With regard to an analysis using an anti-MUC18 antibody, US 6,184,043 B1 teaches the metastatic prostate cancer cell line DU-145 does *not* express the MUC18, in obvious contrast to the teachings of the specification (Table 2a). However, an alternative explanation of the results, which is more consistent with the teachings of the specification, is that the antibody made in response to MUC18, and which is used by Fodstad, et al (US 6,184,043 B1), is incapable of detecting metastatic prostate cancer cells. Clearly, there is a high level of unpredictability in the art: two independent investigators unexpectedly found that antibodies made in experimental laboratory animals in response to a MUC18 antigen were incapable of detecting malignant prostate cancer or melanoma cells. In light of the teachings of the references cited above, certainly the skilled artisan cannot predict whether an anti-MUC18 antibody can be used in the claimed method to reliably and accurately identify the metastatic potential of prostate cancer cells. Because the claims embrace an embodiment in which the invention can be used clinically, there is an obvious danger associated with using a method that is not fully reliable and accurate since clearly a misdiagnosis would cause grave consequences to the patient.

Additionally, it is noted that claim 4 does not require that the antibody actually recognize and bind specifically to an epitope of MUC18, only that the antibody be made in response to a MUC18 antigen. If the antibody does not bind specifically to MUC18 or if it cross-reacts with other antigens, and if the cells expressing the cross-reactive antigens are normal and non-metastatic, the identification of metastatic prostate cancer cells will be obviously erroneous or unduly complicated by non-specific staining or background.

In view of the above and particularly in the absence of sufficient guidance with regard to these issues and because of a lack of exemplification that is commensurate in scope to the claims, one skilled in the art cannot practice the invention, when drawn to the full breadth of the claims, with a reasonable expectation of success without undue experimentation.

8. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

9. Claims 1-12 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

In general, claim 1 is not clearly written and it is suggested that the claim be amended so that it reads more comprehensibly.

Claims 1-12 are vague and indefinite because claim 1 recites the term "metastatic potential". The term renders the claim vague and indefinite because the term is not specifically defined in the specification and it cannot be ascertained what specific properties of a prostate cancer cell define the claimed potential for metastasis. It is well known in the relevant that the potential to metastasize can be defined differently. While malignant cancer cells generally have the capacity to metastasize, spreading from the site of primary disease to remote or secondary anatomical sites, there are several indices that the artisan might use to assess this capacity (i.e., potential), including, but not limited to *in vitro* assays measuring the motility of cancer cells, *in vitro* assays measuring the ability of cancer cells to invade subjacent monolayers, and *in vivo* assays in which immune-compromised mice are inoculated with tumor cells and metastases are histologically detected. Therefore, because the meaning of the term "metastatic potential" is not immediately apparent from reading the claim and because the specification does not specifically define the term, one of ordinary skill in the art is not reasonably apprised of the scope of the invention.

Claims 1-12 are indefinite because claim 1 recites the indefinite article *a* in the phrase "a prostate cancer cell" in line 3. The use of the indefinite article renders claim 1 indefinite because it cannot be ascertained whether the "prostate cancer cell" referred to in line 3 is the same or a different cell from that referred to in line 1. Additionally, claim 1 recites the indefinite article *a* in the phrase "a MUC18 coding sequence" in lines 4-5 and line 9. Again, the use of the indefinite article renders the claim indefinite because it cannot be ascertained whether the "MUC19 coding sequence" referred to in lines 4-5 and line 9 is the same or a different MUC18 coding sequence from that referred to in

line 3. Claim 1 also recites the indefinite article *a* in the phrase "a prostate cancer cell" in lines 6-7 and in line 8. Again, the use of the indefinite article renders claim 1 indefinite because it cannot be ascertained whether the "prostate cancer cell" referred to in lines 6-7 and line 8 is the same or a different cell from that referred to in line 1. Amending claim 1 to replace the indefinite articles with definite articles, such as "the" or "said" will obviate these rejections.

Claims 1-12 are also indefinite because claim 1 recites the phrase "wherein a higher level of expression of the MUC18 coding sequence is positively correlated with metastatic potential" in lines 5-6. This phrase renders the claim indefinite because it cannot be ascertained in which cell, the prostate cancer cell or the normal prostate cell, a higher level of expression of the MUC18 coding sequence correlates with metastatic potential. Amending the claim in lines 5-6 to recite, for example, the phrase "wherein a higher level of expression of the MUC18 coding sequence in the prostate cancer cell relative to the level of expression in the normal prostate cell is positively correlated with metastatic potential" can obviate this rejection.

Claims 1-5 and 7-12 are indefinite because claims 1, 3-5, 7, and 10 use the designation "MUC18" as the sole means of identifying the claimed coding sequence. The use of laboratory designations only to identify a particular coding sequence renders the claims indefinite because different laboratories may use the same laboratory designations to define completely distinct coding sequences. In the instant case, it is necessary that the claim be amended to include reference to the specific coding sequence that is embraced by the claim (i.e., identify said coding sequence by a sequence identification number), in order that Applicant distinctly claim the subject matter regarded as the invention and because sequence identification numbers are unique identifiers which unambiguously define a given nucleic acid or protein sequence. Amendment of the claims to include the phrase, for example, "the MUC18 coding sequence set forth in SEQ ID NO: 1" can obviate this rejection. For better clarity and less ambiguity, it is also suggested that claim 4 be amended to recite, for example, the phrase "in response to the MUC18 antigen consisting of the amino acid sequence set forth in SEQ ID NO: 2)".

Claim 5 is vague and indefinite because the claim recites the phrase "a middle portion". The phrase "a middle portion" is vague and indefinite because it cannot be ascertained to which portion of MUC18 the claim refers and therefore one of ordinary skill in the art is not reasonably apprised of the metes and bounds of the invention. Amendment of the claim to recite the specific region of the MUC18 polypeptide that is to be used as an immunogen in producing the anti-MUC18 antibody, referencing the exact positions in SEQ ID NO: 2 that delineate the region of the protein referred to as the middle portion, will obviate this rejection.

Claim 6 is indefinite because the claim recites the indefinite article *an* in line 1. The use of the indefinite article renders the claim indefinite because it cannot be ascertained to which amino acid sequence found in the region spanning amino acids 211 and 376 of SEQ ID NO: 2 the claim specifically refers. Amending the claim to replace the indefinite article for a definite article, such as "the" or "said" can obviate this rejection.

Claims 7 and 10 are indefinite because the claims recite the indefinite article *a* in line 1. The use of the indefinite article renders the claim indefinite because it cannot be ascertained whether the "MUC18 coding sequence" referred to is the same or a different MUC18 coding sequence from that referred to in line 1 of the claim 1, from which claims 7 and 10 depend. Amending claims 7 and 10 to replace the indefinite article with a definite article, such as "the" or "said" will obviate these rejections.

Claim 9 is indefinite because the claim recite the indefinite article *a* in the phrase "a primer" line 1. The use of the indefinite article renders the claim indefinite because it cannot be ascertained whether the "primer" referred to is the same or a different primer from that referred to in line 1 of the claim 8, from which claim 9 depends. Amending claim 9 to replace the indefinite article with a definite article, such as "the" or "said" will obviate this rejection.

10. Claims 1-12 are further rejected under 35 U.S.C. 112, second paragraph, as being incomplete for omitting essential steps in claim 1, such omission amounting to a gap between the steps. See MPEP § 2172.01. The omitted steps are:

(a) a step in which the level of expression of the MUC18 coding sequence is measured in both the prostate cancer cell and a normal prostate cell,

(b) a step in which the levels of expression in prostate cancer and normal cells are compared.

It is noted that claim 1 recites a method step in which the expression product of the gene encoding MUC18 is *detected*, but clearly the method requires more than a qualitative analysis of gene expression; actually, the method requires a quantitative analysis. Additionally, it is noted that claim 1 recites a correlative statement that indicates whether upon comparison of the levels of expression of the gene encoding MUC18 in prostate cancer and normal cells, a prostate cancer cell is identified as having the potential to metastasize. However, the claim does not actually include the essential step that requires a comparison be made.

Claim Rejections - 35 USC § 102

11. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

12. Claims 1-5 are rejected under 35 U.S.C. 102(b) as being anticipated by Rubenstein, et al (*Prostate* **14**: 383-388, 1989), as evidenced by Shih, et al (*Cancer Research* **54**: 2514-2520, 1994), Liu, et al (*Hinyokika Kiyo Acta Urologica Japonica* **39**: 439-444, 1993), and the annotation that accompanies the MUC18 amino acid sequence entry (Accession No. P43121) in the Swiss Protein Database (see result 1 of the US-09-653-961-2.rsp search report).

The claims are drawn to a method for determining whether a prostate cancer cell has the potential to metastasize, wherein said method comprises detecting the expression of the MUC18 coding sequence in the prostate cancer cell and identifying the prostate cancer cell as having metastatic potential, determining the level of expression when the level of expression in the prostate cancer cell is higher than the

level of expression in a normal prostate cell (claim 1), wherein the prostate cancer cell is from a biopsy tissue sample from a patient (claim 2), or wherein the expression of the MUC18 coding sequence is determined by immunoassay (claim 3) wherein the immunoassay uses an antibody made in an experimental laboratory animal in response to a MUC18 antigen (claim 4) wherein the antigen is a middle portion of MUC18 (claim 5).

Rubenstein, et al teach an immunohistologic method for assessing the metastatic potential of prostate cancer cells (abstract). Specifically, Rubenstein, et al teach that the method can be used to distinguish benign prostatic tissue and malignant prostatic tissue (i.e., tissue with metastatic potential) (abstract). The method of Rubenstein, et al comprises the detection and enumeration of more than one tumor-associated antigen or marker on the surface of metastatic prostate cells. One of the antigens that is detected in the immunoassay of Rubenstein, et al is also known to be a natural killer (NK) cell marker, which Rubenstein, et al refers to as Leu-7. The Leu-7 antigen is also known as the HNK-1 antigen, as evidenced by Liu, et al. The HNK-1/Leu-7 antigen is actually an epitope of the melanoma-associated antigen A32, which is also known as, and in fact identical to MUC18, as evidenced by Shih, et al and the annotation that accompanies the MUC18 amino acid sequence entry in the Swiss Protein Database (Accession No. P43121). Shih, et al, disclose that the HNK-1/Leu-7 antigenic moiety is a characteristic and intrinsic component of MUC18/A32, which actually serves to identify the glycoprotein. Therefore, the anti-HNK-1 antibody that is used by Rubenstein, et al to identify prostate cancer cells that have the potential to metastasize anticipates the anti-MUC18 antibody that in used in the claimed invention. Rubenstein, et al teach that the level of the MUC18 antigen that is detected in the metastatic prostate cancer cell is higher than the level detected in the non-metastatic (i.e., benign) prostate cell. Therefore, it is apparent that the level of expression of the MUC18 coding sequence is higher in the metastatic cancer cell than it is in the non-metastatic or normal prostate cell. Rubenstein, et al also teach that both paraffin-embedded and frozen biopsied tissue samples from patients can be used in their method to identify malignant prostate cancer. It is further noted that the anti-HNK-1 antibody used by Rubenstein, et al is

produced in an experimental laboratory animal in response to a MUC18 antigen, namely the HNK-1 moiety, which is reasonably considered to be an intrinsic part of a middle portion of MUC18.

It should be noted that "a middle of portion of MUC18" is not defined by the claim and the intended metes and bounds of the claim limitation cannot be ascertained (see the 35 USC§ 112, second paragraph rejection above). Thus, it is reasonable to consider the HNK-1 moiety of MUC18 to be contained in *a* middle portion of MUC18.

Additionally, the method of the prior art comprises the same method steps as claimed in the instant invention, that is, contacting the same population of cells with an anti-MUC18 antibody. Thus, the claimed method is anticipated because the method will inherently lead to identifying the metastatic potential of the prostate cancer cells. See *Ex parte Novitski*, 26 USPQ 1389, BPAI 1993.

All the limitations of the claims are met.

Claim Rejections - 35 USC § 103

13. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

14. Claims 1-12 are rejected under 35 U.S.C. 103(a) as being unpatentable over Rubenstein, et al (*Prostate* **14**: 383-388, 1989) in view of Liu, et al (*Hinyokika Kiyo Acta Urologica Japonica* **39**: 439-444, 1993), Shih, et al (*Cancer Research* **54**: 2514-2520, 1994), US Patent No. 5,807,978 A, and in further view of US Patent No. 6,057,105 A, and as evidenced the annotation that accompanies the MUC18 amino acid sequence entry (Accession No. P43121) in the Swiss Protein Database (see result 1 of the US-09-653-961-2.rsp search report).

The claims are drawn to a method for determining whether a prostate cancer cell has the potential to metastasize, wherein said method comprises detecting the

expression of the MUC18 coding sequence in the prostate cancer cell and identifying the prostate cancer cell as having metastatic potential, determining the level of expression when the level of expression in the prostate cancer cell is higher than the level of expression in a normal prostate cell (claim 1), wherein the prostate cancer cell is from a biopsy tissue sample from a patient (claim 2), or wherein the expression of the MUC18 coding sequence is determined by immunoassay (claim 3) wherein the immunoassay uses an antibody made in an experimental laboratory animal in response to a MUC18 antigen (claim 4) wherein the antigen is a middle portion of MUC18 (claim 5) wherein said middle portion has an amino acid sequence as given in amino acids 211-376 of SEQ ID NO: 2 (claim 6), or wherein the expression of the MUC18 coding sequence is determined by Northern hybridization (claim 7) wherein a probe used comprises at least 15 contiguous nucleotides of SEQ ID NO: 1 (which encodes the MUC18 polypeptide comprising the amino acid sequence set forth in SEQ ID NO: 2) (claim 8), wherein the probe comprises the sequence set forth in either SEQ ID NO: 6, 7, 9, or 10 (claim 9), or wherein the expression of the MUC18 coding sequence is determined by a reverse transcriptase-polymerase chain reaction (claim 10) wherein a primer used comprises the sequence set forth in either SEQ ID NO: 6, 7, 9, or 10 (claim 11), or wherein the prostate cancer cell in a cell line cell (claim 12).

Rubenstein, et al teach that which is set forth in the 35 USC § 102(b) rejection above, but do not disclose the use of an anti-MUC18 antibody that is made in response to the middle portion of the MUC18 polypeptide, which spans from position 211-376 in the amino acid sequence set forth in SEQ ID NO: 2. Rubenstein, et al does not teach the use of a prostate cancer cell line. Furthermore, Rubenstein, et al do not disclose that the expression of MUC18 can be determined by Northern hybridization analysis, wherein a probe comprising at least 15 contiguous nucleotides of SEQ ID NO: 1 is used, or wherein the probe comprises the sequence set forth in either SEQ ID NO: 6, 7, 9, or 10. Also, Rubenstein, et al do not disclose that the expression of MUC18 can be determined by a reverse transcriptase-polymerase chain (RT-PCR) reaction analysis, wherein a primer used in the RT-PCR analysis comprises the sequence set forth in either SEQ ID NO: 6, 7, 9, or 10.

Liu, et al teach that the Leu-7 tumor-associated antigen detected in the immunoassay of Rubenstein, et al is also known as the HNK-1 antigen. Liu, et al also teach that "the expression of the HNK-1 antigen on prostatic cancer may be a useful prognostic factor in patients with prostatic cancer" (abstract). Further, Liu, et al teach that "of the 52 patients with prostatic cancer, 49 patients (94%) showed reactivity to anti-HNK-1 MAb [monoclonal antibody] and the immunoreaction was associated with the histological differentiation of prostatic cancer" (abstract).

Shih, et al teach that the HNK-1/Leu-7 antigen is actually an epitope of the melanoma-associated antigen A32 (abstract). Specifically, Shih, et al disclose that the "the melanoma-associated antigen, A32, was defined by a murine monoclonal antibody and was immunoprecipitated as a single 113 kDa integral membrane glycoprotein containing sialic acid and HNK-1 carbohydrate moieties" (abstract). Shih, et al teach that the A32 antigen showed sequence identity to the MUC18 antigen (abstract), which is also evidenced by the annotation that accompanies the MUC18 amino acid sequence entry in the Swiss Protein Database (Accession No. P43121). Additionally, Shih, et al teach that "MAb HNK-1 reacted with the affinity-purified A32 antigen, indicating the presence of the HNK-1 carbohydrate moiety on the protein" (page 2515, column 2). Shih, et al, further teach that "besides ICAM-1, melanoma cells express three additional members of the immunoglobulin supergene family, *i.e.,* vascular CAM, NCAM, and MUC18 antigen. Of these, only the MUC18 antigen is expressed on most melanoma lesions, and its expression correlates significantly with metastatic potential" (citations omitted) (page 2514, columns 1-2). Also, Shih, et al teach the use of cell lines (page 2514, column 2).

US Patent No. 5,807,978 A teaches a method for identifying peptides derived from prostate tumor-associated antigens that correspond to the immunodominant epitopes found in the native antigens (abstract), which can be used to produce reagent antibodies that are capable of specifically binding said antigens for use in diagnostic assays (column 3, lines 41-45). US 5,807,978 A teaches that the rationale for using the invention is that "in light of the structural relationship between PSA [prostate-specific antigen] and other molecules, however, it appears that it will be necessary to generate

antibodies against fragments of PSA, rather than against the entire molecule" (column 1, lines 60-64). US 5,807,978 A also teaches that "in theory the application of the immunological model described above could be applied to practically any polypeptide" (column 7, lines 3-5) and as an example, teaches that the invention can be used to generate an anti-MUC18 antibody (column 7, line 29).

US Patent No. 6,057,105 A teaches methods for detecting melanoma cells that have metastatic potential (abstract). The method of US 6,057,105 A comprises detecting the level of expression of the gene encoding MUC18 (column 6, lines 15-21) by quantitative RT-PCR analysis (column 3, line 55 – column 4, line 44), in which a primer of at least 15 contiguous nucleotides of SEQ ID NO: 1 (i.e., the MUC18 coding sequence) is used (column 8, line 42 – column 9, line 33 and column 15, line 48 – column 16, line 5).

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to identify, according to the method of US 5,807,978 A, the middle portion of the MUC18 polypeptide, which spans from position 211 to 376 of the amino acid sequence set forth in SEQ ID NO: 2, that corresponds to an immunodominant epitope found in the native MUC18 polypeptide and then to produce an anti-MUC18 antibody in a laboratory animal in response to the peptide immunogen. Furthermore, it would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to use the anti-MUC18 antibody in the method of Rubenstein, et al to identify prostate cancer cells with metastatic potential, because the anti-MUC18 antibody made according to the method of US 5,807,978 A can be used in parallel experiments to confirm the results of the immunoassay, in which the anti-HNK-1 antibody is used to measure the level of expression of the MUC18 coding sequence in prostate cancer cells and also because Liu, et al teach that the level of expression of the HNK-1/Leu-7 epitope of MUC18/A32 is a valuable prognostic factor associated with prostate cancer cell differentiation. One of ordinary skill in the art at the time the invention was made would have been motivated to make and use the anti-MUC18 antibody, according to the methods of US 5,807,978 A and Rubenstein, et al, respectively, because confirmation of the data acquired in the immunoassay using anti-

HNK-1 antibody would enable a more accurate assessment of the metastatic potential of a patient's prostate cancer cell and thereby increase the ability of the clinician to successfully intervene in the progress of the disease in the patient.

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to use prostate cancer cell lines as positive (e.g., PC-3) and negative controls (e.g., LNCaP) in the method for identifying prostate cancer cells isolated from patient biopsies, because, although in the art it is conventional to use cell lines as controls in experiments, Shih, et al also teach that cell lines can be used. One of ordinary skill in the art at the time the invention was made would have been motivated to use prostate cancer cell lines as controls in the method because it is desirable to identify the frequency of false negative or false positive test results in order to estimate the accuracy of the results.

Additionally, it would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to substitute the RT-PCR method of US 6,057,105 A for the immunoassay of Rubenstein, et al and/or US 5,807,978 A to identify the metastatic potential of prostate cancer cells, because the RT-PCR assay is very sensitive and can be used to detect low levels of expression of the MUC18 coding sequence. It would also have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to use Northern analysis, rather than RT-PCR as a substitute for the immunoassay of Rubenstein, et al and/or US 5,807,978 A to identify the metastatic potential of prostate cancer cells, because it is well known that Northern analysis is a conventional method than can be used alternatively to RT-PCR to determine the levels of expression of the gene encoding MUC18 in prostate cancer cells. It would also have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to use one of the polynucleotides comprising either the sequence set forth in SEQ ID NO: 6, 7, 9, or 10 as a primer in RT-PCR or as a probe in Northern hybridization, because it obvious to one of ordinary skill to derive a primer or probe of at least 15 contiguous nucleotides from the sequence of the gene of interest, in order to study the level of expression of said gene, which in this instance is the MUC18 coding sequence comprising the polynucleotide sequence set forth in SEQ ID NO: 1.

One of ordinary skill in the art at the time the invention was made would have been motivated to use either RT-PCR or Northern hybridization to confirm the results of the immunoassay of Rubenstein, et al, and/or US 5,807978 A, because confirmation of the data acquired in the immunoassay by a different method would enable a more accurate assessment of the metastatic potential of a patient's prostate cancer cell and thereby increase the ability of the clinician to successfully intervene in the progress of the disease in the patient.

Conclusion

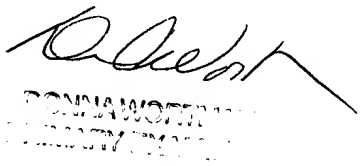
15. No claims are allowed.

16. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Stephen L. Rawlings, Ph.D. whose telephone number is (703) 305-3008. The examiner can normally be reached on Monday-Thursday, alternate Fridays, 8:00AM-5:30PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Anthony C. Caputa, Ph.D. can be reached on (703) 308-3995. The fax phone numbers for the organization where this application or proceeding is assigned are (703) 308-4242 for regular communications and (703) 308-4242 for After Final communications.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (703) 308-0196.

Stephen L. Rawlings, Ph.D.
Art Unit 1642



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April 5, 2001